201-14506

NCIC HPV
Sent by: Mary-Beth Weaver

05/30/2003 08:58 AM

To: NCIC HPV, moran.matthew@epa.gov
cc:
cc:
Subject: Public comments on the Ferro HPV test plan for isodecyl diphenyl phosphate

Jessica Sandler <jessicas@peta.org> on 05/28/2003 12:36:24 PM

To: olsona@ferro.com, oppt.ncic@epamail.epa.gov, hpv.chemrtk@epamail.epa.gov, Rtk Chem/DC/USEPA/US@EPA, Karen Boswell/DC/USEPA/US@EPA
cc: Priscilla Flattery/DC/USEPA/US@EPA, Oscar Hernandez/DC/USEPA/US@EPA, Stephen Johnson/DC/USEPA/US@EPA

Subject: Public comments on the Ferro HPV test plan for isodecyl diphenyl phosphate

Dear Mr. Johnson,

Attached please find the comments of the American animal protection community on Ferro Corporation's HPV test plan for isodecyl diphenyl phosphate. I am calling your attention to it as it is particularly egregious. We are asking that you review our comments prior to issuing your own and that you address the fact that a large amount of existing data has been ignored by the company while proposing to kill large numbers of animals (including in the OECD 414) as well as the other concerns detailed in our comments.

Thank you,

Jessica Sandler, MHS
Federal Agency Liaison
People for the Ethical Treatment of Animals
757-622-7382 ext. 1304
jessicas@peta.org

www.peta.org

HPV test plan comments -- (Ferro) Isodecyl diphenyl phosphate.doc

PEOPLE FOR THE ETHICAL TREATMENT OF ANIMALS
501 FRONT STREET
NORFOLK, VA 23510
TEL 757-622-PETA
FAX 757-622-0457

May 28, 2003

Christine Todd Whitman, Administrator
US Environmental Protection Agency
Ariel Rios Building
Room 3000, #1101-A
1200 Pennsylvania Avenue, NW
Washington, DC 20460

Subject: Comments on the HPV test plan for isodecyl diphenyl phosphate

Dear Administrator Whitman:

The following are comments on the test plan for isodecyl diphenyl phosphate (CAS no. 29761-21-5) for the HPV program, submitted by Ferro Corporation. These comments are submitted on behalf of People for the Ethical Treatment of Animals (PETA), the Physicians Committee for Responsible Medicine (PCRM), the Humane Society of the United States, the Doris Day Animal League, and Earth Island Institute. These animal protection, health, and environmental organizations have a combined membership of more than ten million Americans.

This test plan violates both the October 1999 agreement to reduce the number of animals killed in the HPV program and the original HPV framework agreement to which all participants subscribed, in that it ignores existing data while proposing to kill more than 1,300 mammals and 40-120 fish. Ferro is planning to conduct an acute fish toxicity test (OECD no. 203), a mammalian acute toxicity test (OECD no. 425), and a mammalian developmental toxicity test (OECD no. 414). Yet judging from the large amount of data that we found simply by a cursory examination of several databases – with very little effort – it appears that Ferro was unwilling to spend the time and effort necessary to prepare a comprehensive test plan.

It is egregious that Appendix 1 of Ferro's test plan (the "summaries") refers to only two previous toxicity studies (an *in vitro* genotoxicity test using murine lymphoma cells; and a 90-day oral toxicity study in rats), yet numerous studies on the toxicity of isodecyl diphenyl phosphate have been carried out previously. The data from several studies have been published, as detailed below. In addition, the data from at least 55 corporate studies have been submitted to the EPA. The EPA submissions, listed at the beginning of the references, would be available to Ferro under the Freedom of Information Act, and the EPA clearly has access to them. The test plan provides no explanation as to why Ferro has disregarded almost all available data.

The published data include the following:

(i) *Acute oral toxicity in rats.* The conclusion was that the LD_{50} value is above 15.8 grams per kilogram of body weight (g/kg), the maximum dose tested (Johannsen 1977).

(ii) *Acute oral toxicity in domestic fowl.* The conclusion was that the LD_{50} value is above 10.0 g/kg, the maximum dose tested (Johannsen 1977).

(iii) *Subchronic oral toxicity in rats*. Pregnant female rats were administered the compound for 14 days at 300-3,000 mg/kg/day. Toxicity was found at 1,000 mg/kg/day and higher (Robinson 1986).

(iv) *Acute dermal toxicity in rabbits*. The conclusion was that the LD_{50} value is above 5.0 g/kg, the maximum dose tested (Johannsen 1977).

(v) *Female reproductive toxicity in rats.* No reproductive parameters were affected by doses of up to 3,000 mg/kg/day (Robinson 1983).

(vi) *Developmental toxicity in rats.* The conclusion from two studies was that there was no developmental toxicity at doses of up to 3,000 mg/kg/day. Dwarfism was increased at a dose of 1,000 mg/kg/day, but it was considered that this was not due to isodecyl diphenyl phosphate (Robinson 1983, 1986).

(vii) *Neurotoxicity in domestic fowl*. The conclusion was that there was no neurotoxicity at cumulative oral doses of up to 120 g/kg, the maximum dose tested (Johannsen 1977).

(viii) *Acute toxicity in freshwater fish*. The 96-hour LC_{50} value of *Lepomis macrochirus* (bluegill sunfish) at 23 C was calculated to be 6,700 ppm (Dawson 1977).

(ix) *Acute toxicity in sea fish*. The 96-hour LC_{50} value of *Menidia beryllina* (tidewater silverside) at 23 C was calculated to be 1,400 ppm (Dawson 1977).

(x) *Ames test.* No mutagenicity was shown with four *Salmonella typhimurium* strains at 100-10,000 ì g/plate (Zeiger 1987).

The incompleteness of this test plan makes it difficult to critique. We therefore urge the EPA to require Ferro to prepare and resubmit a complete test plan. Therefore, the following criticisms of the test plan as it currently stands are merely provisional:

1. Mammalian acute toxicity test

Clearly animal data are already available and no more animals should be poisoned in acute toxicity tests for this substance. As detailed above, acute oral toxicity data for rats and domestic fowl have been published, and unpublished data are also available to the company and to the EPA. Ferro displays a fundamental lack of concern about animals' lives in that it does not discuss these studies, but simply plans to conduct further similar studies. The published data show that in rats and fowl at least, isodecyl diphenyl phosphate has very low acute toxicity.

In addition, information on an *in vitro* method for testing acute toxicity is available in the Appendix. EPA recommends this test be conducted prior to the use of any *in vivo* acute toxicity test. If, as suspected, the *in vitro* test

demonstrates the substance's lack of toxicity, that information combined with the other existing evidence, should be sufficient to convince the EPA that no more animals should die in acute toxicity tests for this substance. Ferro should pursue this issue directly with the EPA as this appears to be a clear case in which the *in vitro* test could substitute for the *in vivo* test.

2. Mammalian developmental toxicity test

Again, as detailed above, animal data are already available for this endpoint. Data for developmental toxicity in rats have been published, and unpublished data are also available to the company and to the EPA.

Second, the assessment and reduction of hazards to humans should be given higher priority than the generation of theoretical data on animals, especially given the high interspecies variability with compounds of this nature. The test plan provides little information about the use of isodecyl diphenyl phosphate and human exposure to this compound. It merely states that the compound "is a flame retardant for most commercial resins including polyvinyl chloride and its copolymers, polyvinyl acetate and acrylics" (p. 2); in the absence of basic exposure data it is highly premature to plan large-scale tests. The toxicity data available show that isodecyl diphenyl phosphate has very low developmental toxicity in rats. However, rat data are unlikely to be directly applicable to humans, because there are major interspecies differences in developmental toxicity with compounds of this type ("The rat embryo seems to be less susceptible to OP [organophosphorus] compounds than the mouse embryo"; Kitos 1992, p. 396), and further studies on rats will be of little value. Exposure and epidemiology studies are therefore appropriate. If the data obtained suggest that there is cause for concern then, assuming that the aim is to reduce real-world hazards rather than to obtain theoretical data, priority should be given to technical and legislative approaches to exposure reduction, rather than to additional animal data generation.

Third, an *in vitro* method for testing developmental toxicity is available (see Appendix).

Finally, even if an *in vivo* developmental toxicity test were required, Ferro again demonstrates a complete lack of concern for animal welfare by proposing to conduct the OECD no. 414, which will kill at least 1,300 animals, when OECD nos. 421 and 422, which would kill half that number, are available and recommended.

3. Acute fish test

(a) The partition coefficient of isodecyl diphenyl phosphate is too high. Ferro proposes determining the partition coefficient (p. 3). However, the log $K_{o/w}$ value is already known to be 5.44 (Saeger 1979), and the EPA has clearly stated that acute fish tests are inappropriate for compounds with log $K_{o/w}$ values above 4.2. The EPA recommends that with such highly hydrophobic compounds a chronic *Daphnia* test be used instead of acute fish and *Daphnia* tests (EPA *Federal Register,* December 2000, p. 81695).

The uselessness of an additional fish test for isodecyl diphenyl phosphate is supported by its very low aqueous solubility (0.75 ppm; Saeger 1979).

(b) *Several fish tests have already been carried out.* Acute fish toxicity data have been published for both freshwater and marine species (Dawson 1977), and unpublished data from several corporate studies have been submitted to the EPA.

(c) *The ecologic significance of fish tests should be taken into consideration.* Ecotoxicity and mammalian toxicity tests have different purposes: mammalian tests are assumed to be useful for predicting toxicity in individual humans, whereas fish tests are not for predicting toxicity in individual fish, but for predicting economic loss (to commercial and "sport" fisheries) and ecologic damage (fish are an important part of the food chain). The fish test therefore aims to show whether exposure to isodecyl diphenyl phosphate will result in large-scale fish death. However, water pollution can wipe out fish stocks even with no direct toxicity, because killing the food of the fish will lead to starvation. Carps and catfishes are herbivorous, eating mostly algae, whereas most other familiar North American freshwater fish species are carnivorous, eating worms, small crustaceans, smaller fish, insect larvae, etc. However, the toxicity of isodecyl diphenyl phosphate towards these types of organism is unknown, as shown by the inclusion in the test plan of tests on aquatic invertebrates and algae (p. 3). Fish tests should not be carried out while other types of aquatic toxicity are uncertain.

(d) *Several* in vitro *and* in silico *alternatives are available*. See Appendix.

Finally, we must reiterate a number of points made by the EPA in its October 1999 letter to HPV program participants (EPA 1999):

1. In analyzing the adequacy of existing data, participants shall conduct a thoughtful, qualitative analysis rather than use a rote checklist approach. Participants may conclude that there is sufficient data, given the totality of what is known about a chemical, including human experience, that certain endpoints need not be tested.

2. Participants shall maximize the use of existing and scientifically adequate data to minimize further testing.

8. … As with all chemicals, before generating new information, participants should further consider whether any additional information obtained would be useful or relevant.

Ferro's test plan is not only a blatant violation of the above October 1999 letter, but of the original HPV framework agreement to review and submit existing data. Once again, we urge the EPA to reject this plan, and to require the preparation and resubmission of a satisfactorily researched test plan.

Thank you for your attention to these comments. We can be reached via e-mail at RichardT@PETA.org.

Sincerely,

Jessica Sandler, MHS
Federal Agency Liaison
People for the Ethical Treatment of Animals

Richard Thornhill, PhD
Research Associate
PETA Research and Education Foundation

Appendix: *In vitro* and *in silico* test methods

1. In silico *fish test substitute*. Quantitative structure activity relationship (QSAR) programs provide *in silico* methods for estimating toxicity to fish and other aquatic organisms. The EPA itself encourages the use of one established QSAR: ECOSAR (EPA 2002).

2. In vitro *fish test substitutes*:

   (i) TETRATOX is an assay based on the protozoan *Tetrahymena pyriformis* (Larsen 1997). With 50% growth impairment as the endpoint, the results of this assay show close similarity to toxicity in the fathead minnow (Schultz 1997), and the extensive available information demonstrates that TETRATOX is an effective alternative to fish testing. It is in fact already used extensively in industry, and is being considered for regulatory acceptance by the OECD. It is also rapid, easy to use, and inexpensive. On October 23, 2001, PETA and PCRM held a meeting with EPA to facilitate incorporation of an *in vitro* aquatic toxicity test into the HPV program, and Dr. Schultz (Professor of Predictive Toxicology, University of Tennessee College of Veterinary Medicine) made a presentation about TETRATOX. On December 5, 2001, PCRM scientist Nicole Cardello presented the details of this meeting, and our proposal, in a letter to EPA Assistant Administrator Stephen Johnson. After more than one year, there has still been no response from Mr. Johnson or anyone else in the agency. We again request a thoughtful, scientific and specific reply to this letter. It is the stated goal of the EPA to incorporate *in vitro* methods into the HPV program, and this presents an ideal opportunity for action rather than words.

   (ii) The test protocol and performance parameters of the recently validated *Dar*T test are described in detail in Schulte (1994) and Nagel (1998). Briefly, however, it uses fertilized zebrafish (*Danio rerio*) eggs as a surrogate for living fish. The exposure period is 48 hours, and assessed endpoints include coagulation, blastula development, gastrulation, termination of gastrulation, development of somites, movement, tail extension, eye development, circulation, heart rate, pigmentation and edema. Endpoints comparable to *in vivo* lethality include failure to

complete gastrulation after 12 hours, absence of somites after 16 hours, absence of heartbeat after 48 hours, and coagulated eggs. The other endpoints provide further insight for a more detailed assessment of test substances. The reliability and relevance of the *Dar*T test have recently been confirmed in an international validation study coordinated and financed by the German Environmental Protection Agency, and predictions of acute toxicity from the *Dar*T test were highly concordant with *in vivo* reference data (Schulte 1996). This *in vitro* test has been accepted in Germany as a replacement for the use of fish in the assessment of wastewater effluent (Friccius 1995), and is clearly suitable for immediate use as a replacement for the use of fish in the HPV program's screening-level toxicity studies.

3. *Mammalian acute toxicity test substitute*. The test plan states that the acute toxicity test will be "possibly supplemented by *in vitro* testing for dose-range finding" (p. 3). We welcome Ferro's intention to use the *in vitro* cytotoxicity test as an adjunct, but we urge it to discuss with the EPA the possibility of using it as an alternative to the *in vivo* test, particularly given the existing data on this substance. In the Multicentre Evaluation of *In Vitro* Cytotoxicity, a worldwide study organized by the Scandinavian Society for Cell Toxicology, basal cytotoxicity assays were found to be more reliable predictors of human lethal doses, for 50 reference chemicals, than were rodent LD_{50} values (Clemedson 1996a, 1996b, 1998a, 1998b, 2000, Ekwall 1998a, 1998b, 2000). Furthermore, when certain other human toxicokinetic data, such as blood-brain barrier passage and timing of lethal action, were used in conjunction with the cytotoxicity results, the prediction of human lethal concentrations improved markedly (Ekwall 2000). The assay used involves measuring the effects of compounds on the viability of human basal keratinocytes, which is determined from the intensity of staining by neutral red, a dye that is taken up by healthy cells more than by dead and low-viability cells.

4. *Mammalian developmental toxicity test substitute*. An *in vitro* embryotoxicity test method, the rodent embryonic stem cell test, has recently been validated by the European Centre for the Validation of Alternative Methods, and the Centre's Scientific Advisory Committee has concluded that this test is ready to be considered for regulatory purposes (Genschow 2002). This test is now commercially available in the U.S. We therefore urge Ferro to consider the use of this *in vitro* test. If a positive result is found in the embryonic stem cell test, isodecyl diphenyl phosphate should be treated as a developmental toxicant/teratogen, and no further testing should then be carried out within the screening-level program. Although we have written to the EPA repeatedly concerning the inclusion of the embryonic stem cell test in the HPV Program, with correspondence dating back more than eight months, we have received no reply. We urge Ferro to correspond directly with the EPA on the incorporation of this validated non-animal test.

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878211420, 878211421, 878211422, 878211423, 878211567, 878211568, 878211570, 878211571, 878211573, 878211574, 878211575, 878211576, 878211580, 878211581, 878211584, 878211730, 878211881, 878211882, 878211958, 878211959, 878211960, 878211961, 878211962, 878211964, 878211965, 878211966, 878211967, 878213757, 878213908, 40-0042259, 40-6642740, 40-6842745, 40-6842746, 40-6942747, 40-7142753, 40-7742198, 40-7842519, 40-7842520, 40-7942802, 40-8142725, 40-8242832, 40-8342505, 40-8442491, 40-8442509, 40-79421157.

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